## **TITLE:** PHENOTYPIC INVESTIGATION OF CARBAPENEMASE PRODUCTION IN ESBL-POSITIVE AND NEGATIVE *Escherichia coli* ISOLATED FROM A DOMESTIC SEWAGE.

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## **ABSTRACT:**

Escherichia coli, a Gram-negative bacteria of the Enterobacteriales order, has acquired resistance to  $\beta$ -lactam carbapenems, which limits treatment options for infections. Among the mechanisms of resistance to carbapenems, inactivation by the beta-lactamases enzymes has a great impact due to its potential for dissemination. The most common carbapenemases are classified into classes A (e.g. Klebsiella pneumoniae carbapenemase, KPC), B (e.g. New Delhi-beta-lactamase, NDM and Imipenemase, IPM) and D (e.g. Oxacillinases, OXA48 and 181) from Ambler and can inactivate all beta-lactams. Although genotypic tests are considered gold standard in the detection of these enzymes, phenotypic tests such as the modified Hodge test (MHT) and inactivation of carbapenems (mCIM) gain importance considering their low cost and high sensitivity. Therefore, this study's objective was to investigate the production of carbapenemases in extended-spectrum beta-lactamase enzyme (ESBL)-positive and -negative E. coli isolated from domestic sewage, which is important to increase epidemiological knowledge of the distribution of carbapenemases and the coexistence of these beta-lactam resistance mechanisms. A total of 33 E. coli ESBL-positive and negative were submitted to the MHT and mCIM tests. E. coli ATCC 25922 and a clinical lineage of Klebsiella pneumoniae KPC-positive were used as negative and positive control for the tests, respectively. Here, none of the isolates were considered to be carbapenemase producers. Considering the high sensitivity of MHT for detection of KPC enzyme, it may be suggested that the KPC is probably not circulating among the studied samples or that gene encoding is present in these isolates but with a low level of expression, which requires genotypic confirmation. The negative result in the mCIM test, which has a sensitivity of 100% for detection of class A, B and D carbapenemases, confirmed the finding in MHT. Thus, it is suggested that E. coli from domestic sewage still has lower resistance to carbapenems, and the condition of being ESBL-positive does not seem to be related to the production of carbapenemases.

Keywords: Phenotypic Tests, Enterobacteriales, ESBL, Carbapenemases, Carbapenems.

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